

NOTE

Podite carrying ciliates dominate the benthic ciliate community in the kelp forest

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ABSTRACT: The ciliate fauna on the kelp *Laminaria hyperborea* and 5 of its epiphytes was sampled in April and August, off the north-west coast of Norway. The lamina of the kelp carried few (old lamina) or no ciliates (new lamina), while podite carrying cyrtophorids of the order Dysteriida prevailed in April on the epiphytes. The highest number of ciliates was found on *Ectocarpus* sp. (total ciliate number of 20352 ± 5040 ciliates g^{-1} fresh weight, FW), with species of the genus *Dysteria* (8008 ± 1928 g^{-1} FW) and 1 species of the genus *Hartmannula* (*Hartmannula angustipilosa*; 8767 ± 5202 g^{-1} FW) being dominant. In August, the abundance of ciliates was greatly reduced on all epiphytes, although more taxa were observed. *Aspidisca* spp. (4829 ± 987 g^{-1} FW) dominated total numbers (6080 ± 1118 ciliates g^{-1} FW) on *Ectocarpus* sp. The infraciliature of 7 species from the family Dysteridae, including *Agnathodysteria littoralis*, *Trochilia sigmoides* and *Dysteria monostyla*, is described. The dysterids and *H. angustipilosa* carry a flexible podite by which they attach to the surface. The podite may be the key to success in this wave- and current-exposed environment.

KEY WORDS: Benthic ciliates · *Dysteria* spp. · Cyrtophorids · Kelp forest · Epiphytes · Abundance

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INTRODUCTION

While the importance and function of ciliates in planktonic food webs have received much attention over the last decades (Gismervik et al. 1997, Sherr & Sherr 2002), the role of ciliates associated to benthic algal communities has been explored to a far lesser extent. Kelp forest communities are highly productive ecosystems (Mann 2000). Along the Norwegian coast, these forests are dominated by the large brown algae *Laminaria hyperborea* (Gunnerus) Foslie (Kain 1967), which can reach a length of 3.5 m and an age between 10 and 20 yr (Sjötun et al. 1995). The kelp consists of a holdfast and a stipe, as well as a lamina which is exchanged every year. The stipe is typically overgrown with annual epiphytic algae, and altogether these plants make up a heterogeneous and seasonally changing habitat for a variety of animals (Christie et al. 1998, Norderhaug et al. 2002). Considering the high

production of carbon in the kelp forest, much of which is channelled through the microbial food web by bacteria utilising DOM and POM from ageing plants and debris (Newell & Lucas 1981, Mann 2000), one should expect protists to be abundant in this system. A few studies have assessed the abundance of ciliates in seaweed communities (Armstrong et al. 1999, Hillebrand et al. 2002), but the contribution of different taxonomic groups as well as their role in the food web remains unexplored. However, the studies performed reveal a range of ciliates with different feeding modes and food preferences (Armstrong et al. 1999, Hillebrand et al. 2002).

This study gives the first data on ciliate taxa and abundance in a kelp forest. Benthic ciliates living on the lamina of *Laminaria hyperborea* and 5 of its epiphytes were sampled in spring and early autumn. The dominant species were identified using protargol stains.

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MATERIALS AND METHODS

Algae were collected by scuba divers off the coast of Finnøy (62° 50' N), north-west Norway. The site is semi-exposed to wave action and contains well-developed forests of *Laminaria hyperborea*. The samples were collected at approximately 4 m depth, put in plastic bags with water, sealed and brought to the surface. Three sub-samples of each algae were taken within 1 h, put in brown bottles, to which 50 to 100 ml water was added, and fixed in 2% acid Lugol's (some algae seemed to absorb some of the fixative, and the samples were re-fixed within a few weeks). Ciliates were also studied alive under an inverted microscope, with and without epifluorescence. Water temperature was 4 and 15°C in April and August 2002, respectively. Salinity in these waters is stable and around 34‰.

In the laboratory, bottles with macroalgae suspended in seawater were gently mixed, and 10 or 50 ml was poured in Utermöhl chambers for enumeration under an inverted microscope at 200× magnification. Small rotifers were also counted in these chambers. Finely branched algae (*Ptilota gunneri* and *Ectocarpus* sp.) were washed to remove ciliates that still adhered to the plant, and the wash water was settled in another Utermöhl chamber. Although living *Dysteria* cells attach to surfaces, it was observed that the cells became detached as soon as fixative was added. Sessile ciliates like peritrichs were not collected using this method. Excess water was removed from the algae, and the algal fresh weight (FW) recorded.

Few ciliates were found in Lugol samples containing the lamina of the kelp. An additional method was used to check the surface of the kelp, to see whether any ciliates were still present on the lamina. Formaldehyde fixed sub-samples of the lamina (3.8 cm²) were sub-

merged in DAPI (50 µg ml⁻¹) for 1 min, gently dipped in 0.1 M Trizma-HCl solution and mounted on a slide with paraffin oil. The samples were checked for ciliates at 400× magnification under UV illumination.

Species identification was based on protargol stained samples in accordance with the method of Montagnes & Lynn (1993). The methods of Lee et al. (2000) and Gong et al. (2002) are used for classification and terminology, respectively.

RESULTS AND DISCUSSION

Five different epiphytes with variable surface properties were collected (Table 1). Ciliate numbers were highest on the finely branched algae; in April more than 20 000 ciliates were found on 1 g FW *Ectocarpus* sp. (Table 1). Ciliate abundances on the other epiphytes were 1 to 2 orders of magnitude lower (Table 1). Lowest numbers were found for 2 of the leaf-like algae with a smooth surface. In August, ciliate abundance was reduced on *Ectocarpus* sp. and *Ptilota gunneri* (Fig. 1). Numbers remained approximately the same in both months for the other algae, but there was a change in dominant taxa (Fig. 1). Members of the genus *Dysteria* Huxley, 1857 dominated on all epiphytes in April, except on *Ectocarpus* sp., where the species *Hartmannula angustipilosa* (Deroux & Dragesco 1968) of the genus *Hartmannula* Poche 1913 was dominant, reaching numbers similar to those for *Dysteria* (Fig. 1). In August, numbers of *Dysteria* spp. were in the same range as other taxa, and a small (<20 µm) relative from the genus *Trochilia* appeared (*Trochilia sigmoides* Deroux, 1976). *Aspidisca* spp. became more important in August, and although dominated by *Aspidisca steinii* Buddenbrock, 1920 (Song & Wilbert 1997), a smaller unidentified species

Table 1. List of macroalgal species, period of sampling, mean number of ciliates recorded per gram fresh weight (FW) algae ± SD (n = 3), and dominant taxa of ciliates. nd: no data

Macroalgae	Algae structure	Sampled	Ciliates g ⁻¹ FW	Dominant ciliate taxa
<i>Laminaria hyperborea</i> :				
Old lamina without epiphytes	Leaf-like leathery	April		A few specimens found
New lamina		April		No ciliates found on the lamina
		August		No ciliates found on the lamina
<i>Palmaria palmata</i>	Leaf-like lobed	April	138 ± 17	Small <i>Dysteria</i> sp. (30–35 µm)
		August	136 ± 39	<i>Aspidisca steinii</i>
<i>Delesseria sanguinea</i>	Leaf-like smooth	April	170 ± 178	Small <i>Dysteria</i> sp. (30–35 µm)
		August	215 ± 125	<i>Aspidisca steinii</i>
				Species from subclass Stichotricha
<i>Membranoptera alata</i>	Leaf-like smooth	April	2396 ± 757	Small <i>Dysteria</i> sp. (30–35 µm)
		August	nd	Not sampled
<i>Ptilota gunneri</i>	Branched compressed	April	2120 ± 1038	Small <i>Dysteria</i> sp. (30–35 µm)
		August	331 ± 40	<i>Aspidisca steinii</i>
<i>Ectocarpus</i> sp.	Filamentous finely branched	April	20352 ± 5040	<i>Hartmannula angustipilosa</i>
		August	6080 ± 1118	<i>Aspidisca</i> spp.

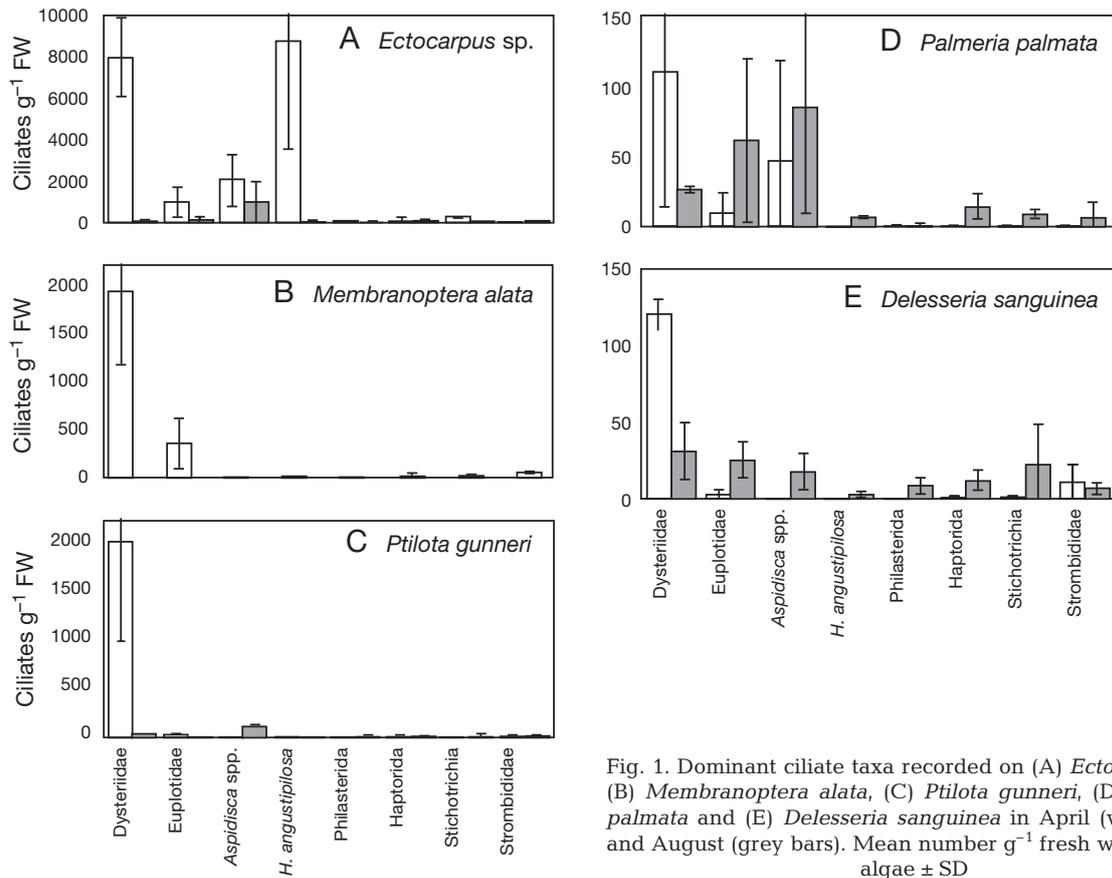


Fig. 1. Dominant ciliate taxa recorded on (A) *Ectocarpus* sp., (B) *Membranoptera alata*, (C) *Ptilota gunneri*, (D) *Palmaria palmata* and (E) *Delesseria sanguinea* in April (white bars) and August (grey bars). Mean number g^{-1} fresh weight (FW) algae \pm SD

was then frequently recorded. *Euplotes* spp. were always present, but never in high numbers. Common species from the order Haptorida were *Dileptus* sp., *Trachelius* sp. and *Lacrymaria* sp.

In contrast to the high numbers of ciliates found on the epiphytes of the kelp, no ciliates were recorded on the lamina itself in August, while a few were observed on old kelp with epiphytes from April (this kelp was from the previous year) (Table 1). Direct observation of

the lamina under epifluorescence (see 'Materials and methods') confirmed the results from the lugol fixed samples. The surface of the kelp was smooth and protists were only present on the old lamina or where the lamina had been damaged.

At least 6 different species from the genus *Dysteria* were found (Fig. 2A–D, Table 2). Individuals were assigned to different categories of size and shape. From these counts, combined with the study of pro-

Table 2. Infraciliature of 7 species from the family Dysteriidae. Size measurements (length, height) are of protargol stained specimens, mean \pm SD; n = number of specimens. RvK: right ventral kinety; LK: left equatorial kinety; FvK: frontoventral kinety (sensu Gong et al. 2002, see also Fig. 2). Numbers in parentheses are number of specimens recorded with clearly visible kineties

Species	Cell size (μ m)	n	RvK	LK	FvK	Habitat	Position of cytopharynx
<i>Trochilia sigmoides</i>	13.0 \pm 0.9, 9.6 \pm 0.9	10	2 (10)	2–4	2 (10)	<i>Ectocarpus</i> sp.	Diagonal
<i>Agnathodysteria littoralis</i>	30.7 \pm 3.5, 18.6 \pm 1.4	10	4 (8)	4–5 (4)	3 (9)	<i>Ptilota gunneri</i> <i>Ectocarpus</i> sp. <i>Palmaria palmata</i>	Vertical/ diagonal
Slender <i>Dysteria</i> sp.	33, 16	1	3	>4	1	<i>Ectocarpus</i> sp.	Longitudinal
Medium <i>Dysteria</i> sp. 1	36.7 \pm 3.7, 19.2 \pm 1.8	10	4 (10)	3–5 (4)	1 (10)	<i>Ptilota gunneri</i> <i>Ectocarpus</i> sp.	Diagonal
Medium <i>Dysteria</i> sp. 2	36.7 \pm 5.4, 22.5 \pm 3.3	6	6–7 (5)	4 (2)	1 (5)	<i>Ptilota gunneri</i>	Diagonal
<i>Dysteria monostyla</i>	39.0 \pm 2.8, 25.5 \pm 2.7	2	4 (2)	Yes	2 (2)	<i>Ectocarpus</i> sp.	Longitudinal
Large <i>Dysteria</i> sp.	84, 56	1	7	7	3	<i>Delesseria sanguinea</i>	Longitudinal

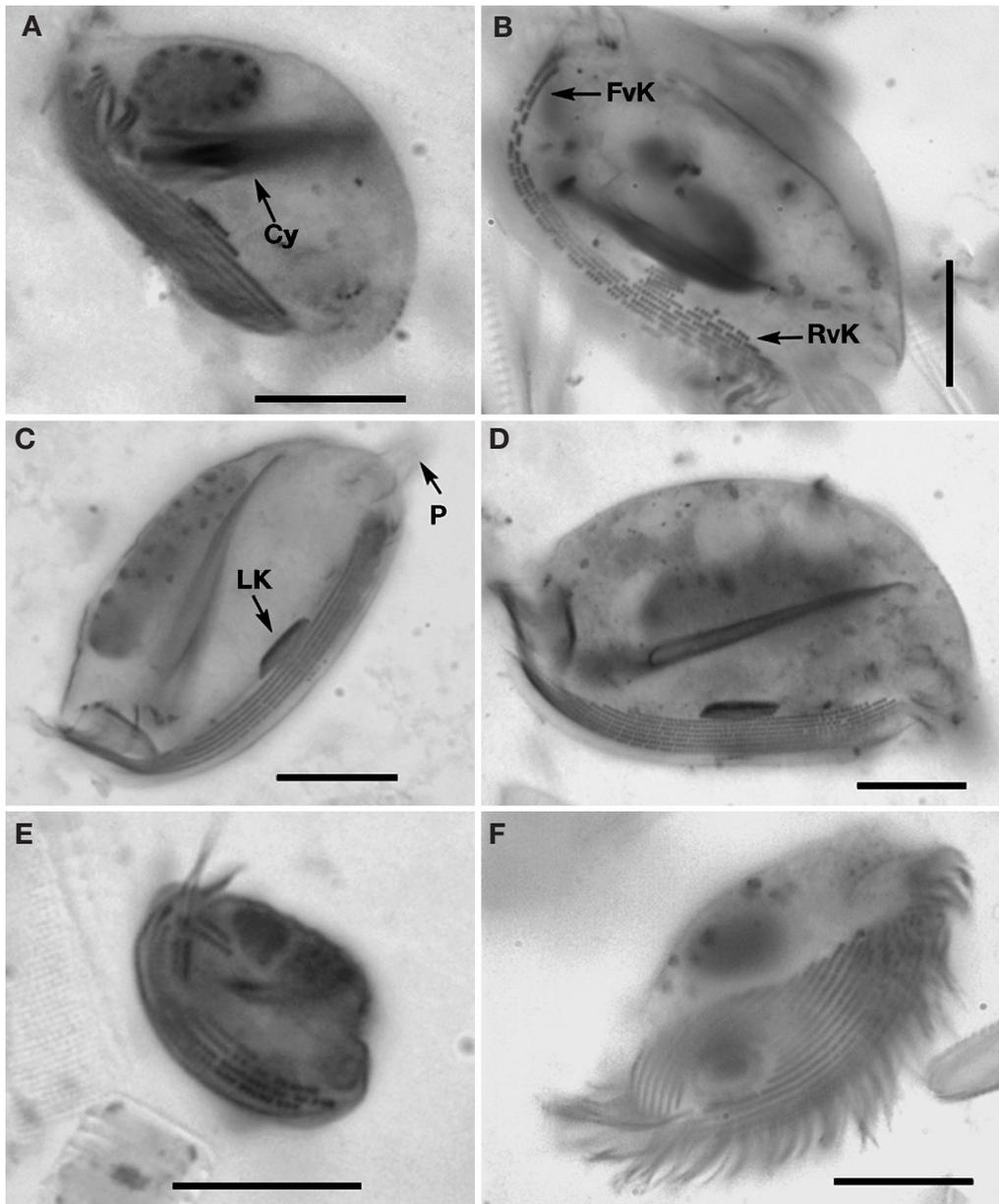


Fig. 2. Protargol stained specimens of (A) *Agnathodysteria littoralis*, (B) *Dysteria monostyla*, (C) *Dysteria* sp. 1, (D) *Dysteria* sp. 2, (E) *Trochilia sigmoides*, and (F) *Hartmannula angustipilosa*. Scale bars = 10 μ m. Cy: cytopharynx; FvK: frontoventral kinety; RvK: right ventral kinety; LK: left equatorial kinety; P: podite

targol stained specimens, it was concluded that *Agnathodysteria littoralis* Deroux, 1976 dominated by number. To my knowledge, this ciliate has only been reported by Deroux (1976), who collected the species from the brown algae *Fucus serratus*. It was considered common, but never abundant. It is difficult to say whether this species has been described previously, as the infraciliature of dysterids was not reported in earlier taxonomic works of, e.g., Kahl (1931).

Four other medium sized *Dysteria* species (37–39 μ m long) were frequently recorded while 1 large species

(ca. >80 μ m) was rare (Table 2). The species were identified on the basis of their ventral infraciliature, which is highly species specific (Gong et al. 2002), cell size and position of cytopharynx (Table 2). Although about 30 nominal species within the genus *Dysteria* have been described, the infraciliature is only known for 7 of these (see Gong et al. 2002 and references therein). The most frequently reported species, *Dysteria monostyla*, is widespread and has been encountered in several regions of the Atlantic Ocean as well as in the Yellow Sea (Stein 1859, Deroux 1965,

Dragesco & Dragesco-Kernéis 1986, Gong et al. 2002). The specimens found in this study differ slightly from the previously described forms, as they have 4 rather than 3 right ventral kineties. However, the last kinety of the right ventral kinety is short (Fig. 2), and terminates behind the left equatorial kinety. The species is named *D. monostyla* here, as its other features regarding infraciliature, cytopharynx, size and form are in concert with previous descriptions.

In the Lugol fixed samples, *Hartmannula angustipilosa* was distinguished by its peculiar shape, as the anterior end clearly protruded from the rest of the oval cell (Fig. 2F). This species also carries a small podite, and attaches to the substrate by a thread like *Dysteria* spp. The kelp forest is usually situated in exposed areas, subject to strong tidal currents and wave action. The ability of the dysteriids to attach to the substratum by a thread may explain their success in this particular system.

Live observations and Lugol samples revealed high numbers of a small (<100 µm) rotifer of the genus *Colurella*. In April, nearly 8000 specimens were recorded for 1 g FW *Ectocarpus* sp. (data not shown), while numbers were low for the other macroalgae (<20 ind. g⁻¹ FW), as well as for all algae in August (<300 ind. g⁻¹ FW). With their foot retracted into their lorica, *Colurella* resemble a dysteriid, both in shape and in size, and care should be taken not to mistake these multi-cellular organisms for ciliates (see Turner 1995). According to Thane-Fenchel (1968) these rotifers feed on diatoms, dinoflagellates and various bacteria. The dysterid ciliates (both *Dysteria* and *Hartmannula*) may also exploit single and filamentous bacteria (Fenchel 1987, Gong et al. 2002), and personal observation of dysteriids under epifluorescence suggests that they feed on microalgae. Thus, the small rotifers are likely competitors to ciliates, rather than predators. In spring, the surface of the epiphytes was covered with pennate diatoms. These were observed inside protargol stained cells of both strombidiid ciliates and members of Haptorida, suggesting a flow of carbon from primary producers, using the macrophytes as substrate, to ciliates, in addition to a microbial pathway, from macrophyte carbon via bacteria to protozoans.

More taxa were observed in August than in April, although total numbers declined. Despite variable surface properties, and variable abundance of ciliates on the different epiphytes (100 to 6000 ciliates g⁻¹ FW), the same number of taxa were recorded on all epiphytes in August. A corresponding pattern has been found for macrofauna associated with various macroalgae (Lippert et al. 2001, Christie et al. 2003). This suggests that factors other than surface properties and habitats may play an important part in increasing numbers of taxa, like increased food diversity. Also, in

August the metazoan community was abundant and diverse compared to the situation in spring, indicating increased predation pressure on ciliates (Christie et al. 2003). An increase in bacterivorous (*Aspidisca steinii*, *Uronema* sp.) as well as carnivorous species (*Dileptus* sp., *Lacrymaria* sp.) further suggests that both the food and predator regime had changed compared to the situation in spring. Another factor that may explain the increased diversity is that while the spring community is young and newly established after the winter season, the early autumn community has had time to recruit specimens from other plants and areas. Thus, the diversity and abundance of the ciliate community in the kelp forest results from a balance between establishment of habitat, recruitment of species, and changing food availability on the one hand, and predation pressure from a variety of micro-, meio- and macrofauna on the other hand.

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